

of these prioritisation scenarios, we obtain a priority map, which we compare with each other in order to identify factors to which the prioritisation is most sensitive. This exercise was repeated with different underlying landscapes and alien distributions to assess effects of these latter two factors on the sensitivity of the factors. In a second step, we will determine how selected changes in the prioritisation scenarios impacts on the status of IAPs over a time period of 100 years, with the aim of identifying the most effective prioritisation strategy.

doi:[10.1016/j.sajb.2009.02.072](https://doi.org/10.1016/j.sajb.2009.02.072)

### Antimycobacterial activity of seven herbaceous plants

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Tuberculosis (TB) is the world's longest running catastrophe and even though relatively adequate treatments are in place, emerging multiple drug resistance (MDR) has become a major threat in recent years and thus calls for an urgent search for new and effective treatments for this deadly disease. In the present study, seven herbaceous plants were selected for anti-tuberculous activity based on phytochemical constituents and ethnobotanical information. Ethanol and dichloromethane extracts of all the plants were first screened for antimycobacterial activity against non-pathogenic mycobacterium (*Mycobacterium smegmatis*). The best samples were found to be the aerial ethanolic extracts of, *Knowltonia vesicatoria* and the *Helleborus* (cultivar), which exhibited minimum inhibitory concentrations (MIC) of 0.6 mg/ml and 1.2 mg/ml, respectively and minimum bactericidal concentrations (MBC) of 1.2 mg/ml and 5 mg/ml. Four selected extracts were further tested for anti-tuberculous activity against extracellular *Mycobacterium tuberculosis* (strain H37Rv) using the BACTEC radiometric method. *K. vesicatoria* had the lowest MIC of 0.1 mg/ml and *Symphytum officinale* had the highest MIC of 5 mg/ml whilst both *Helleborus* and *Ranunculus repens* showed MIC values of 1 mg/ml. These samples, except *S. officinale*, were tested for possible enzymatic mechanism of action utilising mycothiol disulfide reductase (Mtr) assays. No inhibitory activity was evident for any of these at the highest concentrations tested (0.1 mg/ml). Cytotoxicity of these extracts was evaluated on U937 macrophages. Fifty percent inhibitory concentration (IC<sub>50</sub>) of *K. vesicatoria*, *Helleborus* cultivar and *R. repens*

were found to be 0.02 mg/ml, 0.1 mg/ml, and 2.1 mg/ml, respectively. Immunological activity of *K. vesicatoria*, measuring cytokine levels with flow cytometry, is currently underway.

doi:[10.1016/j.sajb.2009.02.073](https://doi.org/10.1016/j.sajb.2009.02.073)

### Anti-cancer activity of plant extracts

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Annually cancer is killing more people than acquired immunodeficiency syndrome, malaria and tuberculosis combined. The infrequency of literature of traditionally used plants in South Africa for the treatment of cancer might be due to the complexity of cancer. The symptoms are more often treated rather than the disease. The aims of the study were to determine the general toxicity of the selected plants, the cytotoxicity of the extracts on cancerous cells and non-cancerous cells and to determine the mechanism of action for the most effective plant. Nine southern African plants were selected based on phytochemical constituents for the present study. These plants were evaluated for toxicity using brine shrimp assays and cancer cell lines. Ethanol extracts of leaves of eight plants were prepared except in one case (*Crotalaria agatiflora*) where leaves and flowers both were extracted with ethanol. The pre-screening of the samples was done using brine shrimp bioassays. Out of the ten samples, eight extracts were found to be non-toxic (Lethal concentration, LC<sub>50</sub> > 1 mg/ml) and two extracts showed moderate toxicity. All extracts were tested for anti-cancer activity on cervical epithelial carcinoma cell line. The fifty percent inhibitory concentration was determined using *in vitro* Sodium 3'-[1-(phenylamino-carbonyl)-3,4-tetrazolium]-bis-[4-methoxy-6-nitro] benzene sulfonic acid hydrate (XTT) cytotoxicity assays. Five extracts namely, *Ghomphocarpus fruticosus* subsp. *fruticosus*, *Crotalaria agatiflora* (leaves and flowers), *Euphorbia damarana* and *Podocarpus henkelii* exhibited IC<sub>50</sub> values < 30 µg/ml and were further tested on non-cancerous cells, Green monkey kidney cells (Vero cells). Leaves of *Crotalaria agatiflora* did not show toxicity on these cells. The IC<sub>50</sub> of leaves was found to be 127.2 µg/ml. Approximately 60% of the cells were found viable on exposure to its flower extract at a concentration of 400 µg/ml. Two extracts namely the *C. agatiflora* flower extract and *C. agatiflora* leaf extract which showed selectively less toxicity on non-cancerous cells and good anti-cancer activity were evaluated for their mechanism of action using DNA fragmentation kit. Results showed that the leaf and flower extracts exhibited moderate signs of apoptosis and necrosis in cultured HeLa cells. Further studies in terms of

identification of bioactive principles of this plant and mechanism involved are recommended.

doi:10.1016/j.sajb.2009.02.074

### An evaluation of molecular and anatomical characters in the genus *Crotalaria*

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*Crotalaria* L. is one of the largest genera in tropical Africa and includes 690 species that are mainly distributed in Africa and Madagascar with some radiation to other parts of the southern hemisphere. The genus is easily recognizable by a combination of the strongly beaked keel, inflated pods, hairs on the upper part of the style and the 5+5 anther arrangement. Polhill thoroughly revised the genus in a publication titled “*Crotalaria* in Africa and Madagascar”. He divided the genus into eight sections based on morphological taximetric analyses. No molecular studies have yet been done to further explore infrageneric relationships and to evaluate the existing hypotheses by applying modern molecular systematic and cladistic techniques. The approach followed in this study includes a broad evaluation of morphological patterns in terms of relationships as revealed by molecular systematic studies. As part of a pilot study in search of additional characters used for taxonomic evaluation, petiole, lamina and fruit anatomy was studied. Molecular methods were optimized using small amounts of plant material (less than 20 mg) for DNA extraction with a DNeasy Plant Mini Kit (Qiagen), changing the reaction components and adjusting amplification and cycle sequence thermal profiles. An overview of the genus will be presented, an evaluation of useful anatomical characters as well as a preliminary molecular phylogeny.

doi:10.1016/j.sajb.2009.02.075

### Sex and bugs and starch degradation

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Starch is a carbohydrate that accumulates in many plants, normally as a medium to long term store of sugar. Bacteria also accumulate a storage polymer, glycogen, which is structurally similar to starch. We thought that we could use the similarities

between glycogen and starch to use the bacteria to identify plant genes which can degrade starch. We therefore transformed a plant cDNA library into *E. coli* and identified colonies which no longer accumulate glycogen. This led to the identification of several genes, some of which code for classical starch degradative enzymes, and some of which show homology only to other plant genes with no known function. The analysis of these genes will be discussed.

doi:10.1016/j.sajb.2009.02.076

### New tribal delimitations in African Apiaceae

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The Apiaceae is a large and taxonomically complex family comprising ca. 463 genera and 3 500 species, with a near cosmopolitan distribution. The Eurocentric classification system of Drude (1898) made little provision for the African taxa. Currently, the family is undergoing a renewed global research effort to produce a natural classification system for the family as a whole. Until recently, the comparatively small sub-Saharan African and Madagascan contingent (80 genera and 354 species) has remained largely unstudied. The high incidence of woodiness and other unusual characters, as well as the many isolated and anomalous taxa indicate that the African genera are critical to an understanding of higher order relationships within the family, particularly in the subfamilies Apioideae and Saniculoideae. As natural relationships are hard to predict on the basis of morphological characters alone, anatomical, morphological and molecular sequence data (*trnQ*, *rps16*, *trnK*) of a selection of species and genera were carefully studied and analyzed. In order to accommodate the unique members of the family substantial rearrangements at both the tribal and subfamilial level are proposed. Many of the African taxa occupy an early diverging position and are here referred to as “Protoapioids”. This group can readily be distinguished from the “Euapioids” (previously referred to as “remaining Apioids”) by the presence of scattered druse crystals in the mesocarp. It is proposed that the latter two groups comprise an expanded Apioideae, which can easily be distinguished from the other two subfamilies, the Azorelloideae and Mackinlayoideae, by the absence of rhomboidal crystals, the non-woody endocarp, the lignification of the mesocarp, and the presence of true wings. A new tribal classification system is proposed, in which five new